ample, when blindness could be relieved temporarily by the resultant improvement in the papilledema. It has been remarked that improvement is in direct ratio to the completeness of the exsections and in inverse ratio to the severity of the disease. This statement represents only an approximation to the fact, and there are several factors involved, some of which are still obscure.

White[16] has tersely stated the case as follows: "Although it is not yet established that the results will be permanent, at least there is good reason to believe that the hands of the clock can be set back for a number of years in the majority of younger patients with hypertension, who have not been permitted to reach the stages of advanced degeneration or changes in heart, kidney."

Poppen[17] of the Lahey Clinic, in a series of 100 consecutive cases, reports good results in 47 per cent, fair results in 24 per cent and unsatisfactory in 22. The mortality was 0.5 per cent. This is rather representative of generally recorded statistics, except as regards the mortality figures which have more usually approached 2 to 5 per cent.

(Editor's Note: Part III, including the references, will appear in the next issue of the *Journal.*)

Antireticular Cytotoxic Serum: A Review*

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ACS or antireticular cytotoxic serum first appeared in an American journal in December 1943. It was greeted with considerable enthusiasm by the lay press in the hope that a remedy had been found for many hitherto hopeless diseases. ACS was first introduced in 1936 by Bogomolets and was said to be a valuable adjunct in the treatment of various disease entities.

Since ACS affects the reticulo-endothelial system, the functions of this system should be mentioned. It is composed of reticular and endothelial elements, derived from the mesoderm, which possess the capacity to ingest foreign body particles and bacterial organisms as well as dyes. These cells are found in the lymph nodes, bone marrow, spleen, liver, thymus, adrenal, lung, and the pituitary gland. Under special circumstances, such as inflammation, connective tissue can be converted to embryonal connective tissue from which mononuclear phagocytes having reticulo-endothelial activity can be mobilized.

Functions of the reticulo-endothelial system have been well reviewed by many authors including Jaffe,[1] Perla,[2] Stern[3] and Bogomolets.[4] The system is concerned with the phagocytosis of red blood cells, metabolism of iron, and the formation of bile; with lipoid, carbohydrate, and protein metabolism; water metabolism; immunity and reaction to infection, and the formation of antibodies. There is a relationship between cancer and the state of activity of the reticulo-endothelial system.[3] Since the reticulo-endothelial system has so many functions, derangement of its function can result in many manifestations of disease, and it becomes understandable how one therapeutic agent which specifically influences this system may affect the course of various diseases.

Metchnikoff in 1900 suggested that the aging process was the result of the accumulation of poisons within the body.[5] Bogomolets suggested that the aging process and disease states could be related to the functional deterioration of the reticulo-endothelial system.[5] He further emphasized that if the system could be stimulated by means of specific cytotoxic sera, then the general health would be improved, degenerative disease prevented or delayed, infectious disease more quickly overcome, and life prolonged. He inferred that ACS would merely help the body help itself by stimulating the natural resistive and protective functions of the body.

In the early phases, work was initiated in this country in three major centers: in Cleveland, Ohio, by Dr. Harry Goldblatt; in Galveston, Texas, by Drs. Anigstein and Pomerat; in Los Angeles by Dr. Reuben Straus and his co-workers.[6] Since then other research centers have started similar investigative work.

This review has been divided into three parts, 1) preparation of the serum, 2) study of the serologic properties of the serum, and 3) testing the biologic effect of the serum in test tubes, in animals, and in human patients.

PREPARATION OF THE SERUM

The serum is prepared by removing spleen and bone marrow from cadavers less than twelve hours postmortem. The cleanly removed tissues are weighed in sterile dishes; four parts of spleen to one part bone marrow are minced with scissors and ground in a mortar with five volumes of normal saline solution. This mixture is centrifuged and the cloudy supernatant fluid used as antigen. The antigen is injected into rabbits and goats at intervals of three days for six doses. On the third to fifth day following the last injection the rabbits are bled by cardiac puncture and the goats from the jugular vein. The resultant serum contains the antireticular cytotoxic antibodies. The strength of the immune serum is tested by complement fixation.[6]

EXPERIMENTAL STUDIES IN ANIMALS

The effect of the serum was tested on the rate of healing of experimentally produced fractures in rabbits. This work was done by Dr. Straus and his associates.[7] To make the experiment more stringent, control rabbits were injected with equivalent amounts of nonimmune serum and, in evaluating the effects of ACS, in addition to the roentgenologic examination, the bones were inspected grossly and microscopically and the breaking strength of the fracture side was measured.

One hundred and fifty-six healthy female rabbits were used. Under intravenous nembutal anesthesia the right radius and ulna were fractured, using an osteoclast to secure as much uniformity as possible in the location, cleanliness, and completeness of the fracture. The limbs were then immobolized in Castex. The animals were divided into four groups. Group A received small or stimulating doses of ACS, Group B larger or depressing doses of ACS, and Groups C and D similar small and large doses of normal goat serum. Roentgenograms were made on the seventh, tenth and fourteenth days after operation. On the fourteenth day the animals were sacrificed, the fractured foreleg amputated and stripped of soft tissues. The roentgenograms were read independently by a roentgenologist and an orthopedist, and the amount of healing recorded.

Examination of roentgenograms of these fractured bones reveals that the group of animals that received stimulating doses of ACS healed much more rapidly than the control group, whereas the group of animals that received the depressing doses of ACS showed evidence of delayed healing that was significantly less than that of the controls. Gross inspection of the bones on the fourteenth day revealed those in animals that received the stimulating dose of ACS to be extremely firm and the union strong, while those in animals receiving depressing doses of ACS revealed very weak union at the fracture site.

Microscopic examination of the bones from all groups of animals showed rather uniformly better formation of callus than the other groups. However, there was extreme overlapping in all groups.

To test the firmness of the union, the bones were placed in a specially devised instrument and subjected to continually increasing pressure to determine the weight necessary to refracture the bones through the fracture site. This experiment showed that the mean breaking strength of experimentally produced fractures of the radius and ulna in rabbits treated with stimulating doses of ACS was nearly twice as great as that of the control fractures after fourteen days, and that large depressive doses caused a marked inhibition of union.

CLINICAL EVALUATION OF EFFECT OF ACS ON HEALING OF FRACTURES

Thus having demonstrated a rather definite effect of ACS on the healing of fractures in rabbits, the next logical step was to see if the serum could affect the healing of fractures in human patients. This work was done on the orthopedic service of the Los Angeles County General Hospital by Dr. Vernon Thompson and his associates.[8] One hundred and seventy-two patients with fractures were studied. Of these, 53 were injected with stimulating doses of ACS. Another group of 40 patients was given normal goat or rabbit serum as a control, and a third group of 79 patients served as uninjected controls. The patients were given volumes of ACS or normal serum varying from 0.03 to 0.25 cc. at intervals of three days for a maximum of fourteen doses. The clinical course and the roentgen findings of each patient were studied objectively. Roentgenograms were taken at two-week or monthly intervals.

Since no good roentgenographic criteria exist to evaluate healing objectively, the following study was devised. The roentgenograms were evaluated in four categories from one to four plus healing.

This was applied to both the development of periosteal and endosteal callus. For periosteal healing the minimal visible amount of callus was designated one plus. If the callus bridged the defect at one or more places, it was termed two plus. If the callus was of moderate density and showed a developing bony architecture, it was designated as three plus. If mature callus could be seen, participating in the formation of a new cortex, it was designated as four plus.

For the evaluation of endosteal callus, if the edges of the fracture fragments were hazy, it was one plus; if the fracture line was partially obliterated by the callus it was termed two plus; if partial trabeculation could be seen extending across the fracture line, it was termed three plus; and if the fracture line was completely obliterated by trabeculae and a new cortex was formed, it was four plus.

This study was made on patients from seventeen to ninety-five years of age, and only fractures of the femur, tibia and humerus were studied.

When the estimation of the extent of healing was plotted against the time in chart form, roentgen-ray evaluation of the healing of tibial fractures showed a definite increase in the extent of healing when compared to the controls. To determine whether or not the estimated differences between the two groups were significant, the different values for each week were subjected to statistical analysis. There were at least some statistically significant observations.

When the data on the fractures of the femur were computed, it was also seen that the ACS-treated patients presented a greater degree of healing per unit of time than did the controls.

In the group of patients with intertrochanteric fractures, again, the ACS-treated patients showed a greater degree of healing per unit of time than did the controls, and graphs were made which revealed those differences what were statistically significant. Similar observations were made for the humerus.

The work just described dealt with the extent of bone healing per unit of time. The reverse, the time required for each group to reach the same extent of healing, was also contrasted. For the four types of fractures, there was an increase in the rate of fracture healing for the ACS-treated patients varying from 19.5 per cent to 50 per cent when those values were compared with the average for the controls.

For clinical evaluation, the orthopedic surgeons studied the clinical data on the chart of each patient. This was done without knowledge of whether the patient was receiving ACS or whether he was a control. Here the personal experience of the clinician was important. In terms of that, he was asked to classify the patient as average, good, or better than average, and poor, or worse than average as to the rate of healing. The results of this evaluation were as follows: Of the control cases 56 per cent were classified as average, 31 per cent better than average, and 13 per cent as poor or worse than average. Of the ACS-treated patients only 38 per cent were considered average, and 62 per cent were considered better than average. None was classified as poor.

It was the impression that pain and tenderness disappeared more quickly in the ACS-treated patients than in the controls.

Six patients with delayed union of fracture were studied. The fractures presented little or no healing for a period of several months to one and one-quarter years. After ACS was started, all but one of these patients went on to complete healing within a relatively short time. The one who failed to heal had a complicated fracture which had insufficient immobilization.

Some of the patients in this series were studied from a biochemical standpoint. The blood proteins, calcium, phosphorus and phosphatase were studied every two weeks. No significant differences were observed in either the ACS-treated patients or the controls.

It would seem from this study that ACS in small or stimulating doses induced an increase in the rate and extent of healing of fractured bones in human patients as well as in rabbits. However, the group of patients is still small, and with improved methods of study that have been suggested additional investigation seems necessary for confirmation. It is possible that more significant biochemical studies can be made on the urinary nitrogen and calcium excretion, according to the technics of Howard and his associates, than were secured in the blood studies.

In experiments measured amounts of spleen and bone marrow tissue freshly removed from rabbits and mice were placed in the Warburg apparatus and graded doses of ACS specific for each species were added. These doses had no

effect on the respiration of spleen or bone marrow. It must therefore be concluded that ACS influences cells in a manner other than by interfering with tissue respiration.

We have been able to kill rabbits with relatively small volumes of ACS, 5 to 10 cc. injected intravenously. No specific histologic changes were noted grossly or microscopically. Further experiments are under way to study the effect of long continued injections of ACS in stimulating and depressing doses.

Not all of our experiments have been productive. We have been unable to stimulate or depress the production of antibodies in rabbits against crystalline egg albumin and typhoid bacilli with stimulating or depressing doses of ACS. Furthermore, we could see no demonstrable effect of either stimulating or depressing doses on the course of experimentally produced influenza and poliomyelitis in mice. In our experiments with the Brown-Pearce carcinoma transplanted to the eyes of rabbits and the Boyer carcinoma transplanted subcutaneously in mice we could observe no significant influence of ACS, in either stimulating or depressing doses, on the growth of these tumors.

We must be extremely cautious in what claims can be made for ACS on human patients. Our experience is limited and we have no good measure of the indirect effects of ACS.

Some measure of hope for slight beneficial effects can be observed in our cancer patients. Not one of our patients, however, was cured. In some, a slight to moderate reduction in the size of tumor masses could be seen. This may have been due to the reduction of regional inflammation or the natural course of the disease. Some patients had improved appetites and gained in weight. These people also experienced a reduction in the amount of pain and a decrease in the necessity for narcotics. In this way some of the patients who were completely bedridden could once more get out of bed and for a while become useful members of society.

We have treated a moderately large group of arthritic patients consisting of those having hypertrophic, rheumatoid, and mixed types of arthritis, both early and late stages. We have not yet made a detailed survey of this group of patients. However, it is our impression that a fair number of the patients with chronic rheumatoid arthritis are improved with ACS therapy; the patients with hypertrophic arthritis are less improved, and the greatest improvement is seen in patients with acute rheumatoid arthritis.

Many of our patients who seemed chronically fatigued appeared to develop renewed energy with ACS, to gain in weight, and to experience a feeling of general well-being. Many other conditions have been tried, but in none have striking results been obtained. As far as we can determine, there is no evidence for the claim of an effect on the duration of life of the patient. Experiments of this nature are contemplated in lower animals.

CONCLUSIONS

From the experiments described by other investigators and those that we have performed, it would appear that we are now dealing with a serum which may have a striking benficial value in the treatment of disease processes. This improvement which is induced is nonspecific and will require more clinical experimentation to reach definitive conclusions. We who work with the serum feel that we must stress conservativeness in our claims; otherwise untold harm may result. It is hoped that other research organizations will undertake experiments so that the evaluation of the effect of serum, whether it be bad or good, will be more quickly reached.

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